Claims

1. A method of identifying one or more nucleic acid sequences useful as a biomarker for a disease to be detected, comprising:

(a) identifying one or more nucleic acid sequences that are down-regulated in diseased cells compared to normal cells, wherein the nucleic acid sequences comprise at least one methylated CpG site in a promoter-first exon region;

(b) comparing expression level of the nucleic acid sequences from (a) with expression level of the nucleic acid sequences from (a) that have been demethylated; and

(c) identifying those nucleic acid sequences exhibiting a significant increase in the expression level after demethylation treatment as compared to the expression level of the same nucleic acid sequences in the methylated state.

2. The method of claim 1, wherein the methylation at one or more specific CpG sites of the nucleic acid sequences from (c) in one or more clinical samples obtained from a subject having or suspected of having the disease to be detected, significantly increases over the normal samples.

3. The method of claim 1, wherein the promoter-first exon region spans about 1000 base pairs upstream of the first exon and about 1000 base pairs downstream of the first exon.

4. The method of claim 1, wherein demethylation is accomplished using a bisulfite compound.

5. A method of detecting, the presence or stage of a disease in a subject, comprising:

(a) determining the degree of methylation of one or more CpG sites on nucleic acid sequences in a biological sample obtained from the subject;

(b) determining the presence of, predisposition to, or stage of the disease in the subject based on the degree of methylation.

6. The method of claim 5, wherein the CpG sites are hypermethylated in cells with the disease.

7. The method of claim 5, wherein the disease is cancer.

8. The method of claim 5, wherein the disease is colorectal cancer.

9. The method of claim 5, wherein the nucleic acid sequences are the ones identified by the method of claim 1.

10. The method of claim 5, wherein the nucleic acid sequences are the ones identified by the method of claim 2.

11. The method of claim 5, wherein the nucleic acid sequences comprise a nucleic acid sequence selected from the group consisting of SEQ ID Nos: 1-129.

12. A method of monitoring the onset, progression, or regression of a disease in a subject, comprising:

(a) detecting in a biological sample of the subject at a first point in time, the degree of methylation of one or more CpG sites on nucleic acid sequences, wherein the CpG sites are differentially methylated at different stages of the disease;

(b) repeating step (a) at a subsequent point in time; and

(c) comparing the degree of methylation of the CpG sites in step (a) and (b), wherein a change in the degree of methylation is indicative of disease progression in the subject.

13. The method of claim 12, wherein the CpG sites are hypermethylated in cells with the disease.

14. The method of claim 12, wherein the disease is cancer.

15. The method of claim 12, wherein the disease is colorectal cancer.

16. The method of claim 12, wherein the CpG sites are the ones identified by the method of claim 1.

17. The method of claim 12, wherein the nucleic acid sequences are the ones identified by the method of claim 2.

18. The method of claim 12, wherein the nucleic acid sequences comprise a nucleic acid sequence selected from the group consisting of SEQ ID Nos: 1-129.

19. A method of determining the efficacy of a test compound for inhibiting a disease in a subject, comprising:

(a) detecting in a first biological sample of the subject, the degree of methylation of one or more CpG sites, wherein the sample has not been exposed to the test compound, and wherein the CpG sites are methylated in the disease;

(b) detecting in a second biological sample of the subject, the degree of methylation of the same CpG sites, wherein the sample has been exposed to the test compound; and

(c) comparing the degree of methylation of the CpG sites in step (a) and (b), wherein a decrease in methylation after the sample has been exposed to the test compound, is indicative of the efficacy of the test compound.

20. The method of claim 19, wherein the CpG sites are hypermethylated in cells with the disease.

21. The method of claim 19, wherein the disease is cancer.

22. The method of claim 19, wherein the disease is colorectal cancer.

23. The method of claim 19, wherein the CpG sites are the ones identified by the method of claim 1.

24. The method of claim 19, wherein the nucleic acid sequences are the ones identified by the method of claim 2.

25. The method of claim 19, wherein the nucleic acid sequences comprise a nucleic acid sequence selected from the group consisting of SEQ ID Nos: 1-129.

26. A kit useful for diagnosis, prognosis, staging, monitoring, and therapeutic treatment of a disease, comprising a bisulfite reagent, and one or more nucleic acid molecules comprising at least about 9 consecutive nucleotides in length that is specific for detecting methylation of one or more CpG sites on one or more nucleic acid marker sequences.

27. The kit of claim 26, further comprising one or more primers and probes comprising at least about 9 consecutive nucleotides in length that is specific for detecting the expression levels of said nucleic acid marker sequences.